Supplemental Information for: Patterns, Variability, and Predictors of Urinary Triclosan Concentrations during Pregnancy and Childhood

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Supplemental Methods Detailing Quality Assurance and Quality Control for Minimizing Contamination

Our study employed several methods to reduce the potential for contamination of our samples with exogenous sources of triclosan. First, we followed previously published recommendations for collecting, storing, and processing biospecimens for analysis of environmental chemical biomarkers.[1,2] Although ideally we would have prescreened all collection and storage materials, the original study was not designed to measure triclosan and thus, we did not test the cups or inserts for triclosan. However, we were able to analyze the wipes that we used to wipe children's genitals and did not detect triclosan in them. The fact that we did not detect triclosan in all samples of each shipment analyzed reassured us that contamination from sampling equipment was not present. Further, reported median triclosan concentrations in the various HOME Study datasets are very similar to medians in several other studies based in the United States conducted at the same time.[3-7]

Second, the Centers for Disease Control and Prevention laboratory, where our urine samples were anlayzed is licensed by the Clinical Laboratory Improvement Act (CLIA) of 1988. Analytical measurements are conducted following strict QA/QC guidelines, CLIA guidelines, and frequent proficiency testing. Quality control (QC) procedures are available online at the NHANES website for each release of data (see below). Each analytic batch includes reagent blanks and low- and high-concentration QC materials, which are evaluated using standard statistical probability rules. The table below includes the coefficients of variation (CV) of the QC materials analyzed with NHANES samples for five NHANES cycles that encompassed the time periods during which we analyzed most (if not all) HOME Study samples. The HOME Study maternal urine samples were analyzed between 2007-2009, while child samples from 1-5 years were analyzed 2010-2012, and samples collected at 8 years of age were analyzed in 2015. The CV are <10% in periods that encompass about one year of analyses.

For blanks included in each batch from any study, should any blank have not been "blank," the analyses would have been out of QC, and we would not have released the data. As currently cited in the manuscript, the CDC lab has previously published work regarding lab practices to track method performance for the quantification of analytes such as triclosan (Ye et al. 2013, EHP) and has pioneered such practices.

Finally, we analyzed a subset of urine samples with and without enzymatic deconjugation, and the results suggested that triclosan was mostly conjugated, thus further ruling out external contamination. Taken together, these data do not suggest systematic contamination during collection or processing of the samples.

Table: Average, standard deviation, and coefficients of variation of low and high concentration quality control samples included in analytic runs at the Centers for Disease Control and Prevention from 2005-2014.

Cycle	N	Start date	End date	Mean TCS	STD of TCS	% CV	Website with details on QC methods
2013-2014	67	6/19/14	11/3/15	49.3	2.6	5.2	https://wwwn.cdc.gov/nchs/data/nhanes/2013-2014/labmethods/EPHPP_H_MET.pdf
	67			16.6	1.5	9.0	
2011-2012	66	4/4/12	3/27/13	67.9	2.9	4.2	http://www.cdc.gov/nchs/data/nhanes/nhanes_11_12/EPH_G_met.pdf
	66			22.7	1.2	5.2	
2009-2010	73	7/19/10	6/27/11	25.2	2.4	9.3	http://www.cdc.gov/nchs/data/nhanes/nhanes_09_10/EPH_F_met_phenols_parabens.pdf
	73			65.4	3.9	5.9	
2007-2008	62	5/7/08	2/9/09	25.3	1.2	6.9	http://www.cdc.gov/nchs/data/nhanes/nhanes_07_08/eph_e_met_phenols_parabens.pdf
	61			65.8	3.4	5.2	
2005-2006	66	6/20/06	2/8/07	26.1	2.5	9.6	http://www.cdc.gov/nchs/data/nhanes/nhanes_05_06/eph_d_met_phenols_parabens.pdf
	66			63.5	5.4	8.4	

Table S1. Summary of studies investigating urinary triclosan concentrations in pregnant women.

Author, Publication Year	Region	Sample Size	Time of Sample Collection	Years of Collection	Triclosan (central tendancy) µg/L
Casas 2011	Spain	120	Third trimester	2005-2006	Median: 6.1
Woodruff 2011	US	268	First, second, or third	2003-2004	Geometric mean: 17
Meeker 2013	Puerto Rico	105	18, 22, and 26 weeks of preg.	2010-2012	Median: 26
Philippat 2013	US	71	At time of amniocentesis appointment	2005-2008	Medians: 6.5-16
Bertelsen 2014	Norway	45	17, 23, and 29 weeks of preg.	2007-2008	Median <LOD 95th percentile: 387
Frederiksen 2014	Denmark	565	Not specified	2011-2012	Median<1.0
Mortensen 2014	US	506	Third trimester	2009-2010	Geometric mean: 19
Philippat 2014	France	520	22-29 weeks of preg.	2003-2006	Median: 30
Pycke 2014	US	181	6-9 mo. of pregnancy	2007-2009	Median: 9.2
Arbuckle 2015	Canada	80	6-19 w, 24-28 w, and 32-36 w	2009-2011	Median: 25
Arbuckle 2015	Canada	1,890	First trimester	2008-2011	Median: 8.7
Current Study	US	383	16 and 26 weeks, at birth	2003-2006	Medians: 11-17

Table S2. Summary of studies investigating childhood urinary triclosan concentrations.

Author, Publication Year	Region	Sample Size	Age	Years of Collection	Triclosan (central tendency) µg/L
Calafat 2008	US	314	6-11	2003-2004	Geometric mean: 8.2 (6.2–11)
Teitelbaum 2008	US	35	6-10	2004	Median: 8.5
Calafat 2009	US	54	NICU premature infants	2003	Not reported
Wolff 2010	US	1,151	6-8 years	2004-2007	Adjusted geometric means: 12-32
Casas 2011	Spain	30	4 years	2005-2006	Median: 1.2
Frederiksen 2014	Denmark	129	6-20 years	2007	Medians: <1-4
Larsson 2014	Sweden	98	6-11 years	Not specified	Not reported
Arbuckle 2015	Canada	80 woman and their infants	<1-3 months	2009-2011	Median: 3.9
CDC 2015	US	314 (2003-04) 356 (2005-06) 389 (2007-08) 415 (2009-10)	6-11	2003-2010	8.2 (2003-04) 13 (2005-06) 12 (2007-08) 11 (2009-10)
Philippat 2015	US	90	Average 5.6 (1.4) years	2007-2009	First visit: 6.6 Second visit: 9.7
Current Study	US	279	1-8 years	2003-2014	Medians: 3.6-17

Table S3. Minimum, maximum, and percentiles of unstandardized (ng/mL) and creatinine-standardized (µg triclosan/g creatinine) urinary triclosan concentrations in HOME Study mothers (16 weeks, 26 weeks, birth) and children (ages 1-5 and 8 years).

			Unstandardized triclosan concentrations (ng/mL)					**Creatinine-standardized triclosan concentrations (µg/g)**				
	N	N(%) <LOD[1]	Min	25th	50th	75th	Max	Min	25th	50th	75th	Max
16 week	387	33(8.5)	<LOD	6.3	16.9	58.4	1,985	0.6	8.0	21.2	63.5	1,248
26 week	371	53(13.7)	<LOD	4.9	13.3	48.9	1,657	0.7	6.3	18.1	62.2	1,453
Birth	345	61(15.8)	<LOD	3.5	11.4	36.6	2,013	0.8	6.2	16.4	51.6	1,968
1 Year	281	111(28.7)	<LOD	<LOD	3.6	11.1	189	2.1	11.5	23.3	56.4	671
2 Years	235	55 (14.2)	<LOD	<LOD	6.8	19.1	7,487	2.2	11.7	22.9	67.6	7,714
3 Years	237	31 (8.0)	<LOD	5.4	16.3	51.9	1,680	1.4	12.0	32.1	98.5	3,988
4 Years	172	21 (5.4)	<LOD	6.2	17.3	54.9	751	1.1	12.6	36.8	95.9	1,325
5 Years	203	15 (3.9)	<LOD	6.3	14.6	38.7	515	1.5	10.0	20.7	59.7	667
8 Years	223	16 (4.1)	<LOD	3.7	9.6	34.5	1,610	0.6	4.2	14.2	36.8	2,080

[1]LOD: limit of detection for pregnancy and ages 1-5 years: 2.3 ng/mL; for 8 years: 1.0 ng/mL

Table S4. Intraclass correlation coefficients (ICCs) of maternal (A) and child (B) urinary triclosan concentrations in the HOME study.
(A)

	16-26 w	**26 w-birth**	**16 w-birth**	**16 w-26 w-birth**
N subjects	369	333	344	382
N samples	738	666	688	1,096
Unstandardized/Unadjusted	0.43 (0.36, 0.51)	0.46 (0.38, 0.53)	0.38 (0.31, 0.46)	0.43 (0.37, 0.48)
Creatinine Standardized	0.54 (0.47, 0.60)	0.58 (0.52, 0.64)	0.49 (0.41, 0.57)	0.53 (0.48, 0.58)
Creatinine Adjusted	0.49 (0.40, 0.56)	0.55 (0.49, 0.61)	0.47 (0.39, 0.54)	0.50 (0.45, 0.56)
Creatinine z-Score Adjusted	0.48 (0.41, 0.56)	0.53 (0.46, 0.60)	0.43 (0.35, 0.50)	0.49 (0.43, 0.54)

(B)

	Long-term (all years)	**Short-term (years 1-3)***	**Toddler (years 1-2)**	**Preschool (years 3-4)**	**Years 1-5**	**School-aged (years 5-8)**
N subjects	299	61	199	151	286	169
N samples	1,312	136	398	302	1,084	338
Unstandardized/Unadjusted	0.22 (0.17, 0.27)	0.54 (0.40, 0.67)	0.27 (0.17, 0.37)	0.39 (0.28, 0.49)	0.21 (0.15, 0.26)	0.28 (0.16, 0.39)
Creatinine Standardized	0.28 (0.23, 0.32)	0.59 (0.46, 0.72)	0.32 (0.22, 0.43)	0.47 (0.36, 0.56)	0.28 (0.23, 0.33)	0.31 (0.20, 0.41)
Creatinine Adjusted	0.27 (0.22, 0.32)	0.56 (0.40, 0.70)	0.33 (0.23, 0.44)	0.44 (0.34, 0.54)	0.28 (0.23, 0.33)	0.30 (0.19, 0.40)
Creatinine z-Score Adjusted	0.24 (0.19, 0.29)	0.58 (0.45, 0.71)	0.29 (0.19, 0.40)	0.44 (0.33, 0.53)	0.22 (0.17, 0.27)	0.32 (0.21, 0.42)

*For subset of children with both a clinic and home visit at ages 1-3

Table S5. Pearson correlations between maternal and child urinary triclosan concentrations, unstandardized and creatinine-standardized.

	Unstandardized						Creatinine-Standardized					
	1-yr	2-yr	3-yr	4-yr	5-yr	8-yr	1-yr	2-yr	3-yr	4-yr	5-yr	8-yr
Maternal average 16-26 w	0.15*	0.07	0.04	0.08	0.08	0.03	0.15*	0.19*	0.04	0.32*	0.20*	0.14*
Maternal average 16 w, 26 w, birth	0.14*	0.11	0.05	0.10	0.11	0.05	0.18*	0.19*	0.06	0.38*	0.24*	0.11

*$p<0.05$

Table S6. Geometric mean (GM) and percent difference (% diff.) in HOME Study children's urinary triclosan concentrations at 8 years of age according to parent-reported child personal care product and cosmetic use (N = 211 with non-missing data).

Variable	N (%)	Unadjusted GM (ng/mL)	Unadjusted % diff. (95% CI)	Adjusted GM (ng/mL)	Adjusted % diff. (95% CI)
Makeup use past 24 h					
No	189 (90%)	12	Ref.	13	Ref.
Yes	22 (10%)	11	-15 (-56, 63)	11	-13 (-56, 74)
Type of makeup past 24 h					
None	192 (91.0%)	12	Ref.	12	Ref.
Mascara, eye shadow, eye liner	7 (3.3%)	16	33 (-58, 323)	17	38 (-57, 339)
Lipstick or lip gloss	6 (2.8%)	6	-51 (-86, 71)	6	-47 (-85, 89)
Nail polish	8 (3.8%)	13	7 (-64, 215)	13	4 (-65, 208)
Other (includes: chapstick)	1 (0.5%)	2	-82 (-99, 287)	2	-82 (-99, 276)
Frequency of hand washing					
Never or once per day	14 (6.6%)	4	Ref.	4	Ref.
2-3 times per day	54 (25.6%)	12	256 (137, 435)	10	171 (75, 319)
4-5 times per day	70 (33.2%)	13	259 (151, 414)	13	226 (125, 374)
>5 times per day	73 (34.6%)	15	334 (205, 516)	16	328 (197, 517)

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